



Abdalrahaman, N. et al. (2015) Deficits in trabecular bone microarchitecture in young women with Type 1 diabetes mellitus. *Journal of Bone and Mineral Research*, 30(8), pp. 1386-1393.

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ARTICLE *in* JOURNAL OF BONE AND MINERAL RESEARCH: THE OFFICIAL JOURNAL OF THE AMERICAN SOCIETY FOR BONE AND MINERAL RESEARCH · JANUARY 2015

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Deficits In Trabecular Bone Microarchitecture In Young Women With Type 1

Diabetes Mellitus

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Keywords - Adiposity, Bone Health, Bone Marrow, Glycaemia, MRI

Running Title - Bone health in T1DM

Word Count (excluding abstract, disclosure and acknowledgements) – 3632

References – 50

Illustrations – 4 Figures and 2 Tables

Abstract

Context: The pathophysiological mechanism of increased fractures in young adults with Type 1 Diabetes Mellitus (T1DM) is unclear.

Objective: Case:control study of trabecular bone microarchitecture and vertebral marrow adiposity in young women with T1DM.

Patients & Settings: 30 women with T1DM with a median (range) age of 22.0yrs (16.9, 36.1) attending one outpatient clinic with a median age at diagnosis of 9.7yrs (0.46, 14.8) were compared to 28 age-matched healthy women who acted as controls.

Methods & Main Outcome Measures: Measurements included MRI-based assessment of proximal tibial bone volume/total volume (appBV/TV), trabecular separation (appTb.Sp), vertebral bone marrow adiposity (BMA) and abdominal adipose tissue and biochemical markers of GH/IGF-1 axis (IGF-1, IGFBP3, ALS) and bone turnover.

Results: Median appBV/TV in cases and controls was 0.3 (0.22, 0.37) and 0.33 (0.26, 0.4), respectively ($p=0.018$) and median appTb.Sp in T1DM was 2.59 (2.24, 3.38) and 2.32 (2.03, 2.97), respectively ($p=0.012$). The median appBV/TV was 0.28 (0.22, 0.33) in those cases with retinopathy ($n=15$) compared to 0.33 (0.25, 0.37) in those without retinopathy ($p=0.02$). Although median visceral adipose tissue in cases was higher than in controls at $5,733\text{mm}^3$ (2030, 11,144) and $3,460\text{mm}^3$ (1,808, 6,832), respectively ($p=0.012$), there was no difference in median BMA which was 31.1% (9.9, 59.9) and 26.3% (8.5, 49.8) in cases and controls, respectively ($p=0.2$). Serum IGF-1 and ALS were also lower in cases and the latter showed an inverse association to appTbSp ($r=-0.30$, $p=0.04$).

Conclusion: Detailed MRI studies in young women with childhood-onset T1DM have shown clear deficits in trabecular microarchitecture of the tibia. Underlying pathophysiological mechanisms may include a microvasculopathy.

Abstract Word Count - 261

Introduction

The risk of hip fractures in those with Type 1 diabetes mellitus (T1DM) is reported to be 7–12 times greater (1, 2) and this increased risk is also evident in young adults (3). The process of differentiation of mesenchymal stem cells into either adipocytes or osteoblasts is regulated by a number of growth factors including insulin, oxygen tension and blood flow within the bone marrow (4, 5). T1DM is also associated with abnormalities of the growth hormone (GH)/insulin-like growth factor type 1 (IGF-1) axis with biochemical evidence of GH resistance (6). Growth hormone (GH) and IGF-1 also are important regulators of bone homeostasis and important for the maintenance of bone mass (7) and may also influence body composition and bone marrow adiposity (8). Mouse models of T1DM exhibit increased bone marrow adiposity (BMA), increased adipocyte markers and increased numbers of lipid-dense adipocytes in the bone marrow (9). Childhood and adolescence are critical periods for skeletal development (10) and it is possible that those affected by T1DM at these ages may be especially susceptible.

On dual energy X-Ray absorptiometry (DXA), adults with T1DM do show a reduction in bone mineral density (BMD) Z-score, reported in a meta-analysis at -0.22 at the lumbar spine and -0.37 at the hip (11), but their fracture risk is much higher (1-3,11) than expected for this modest reduction in BMD. Recent advances in magnetic resonance (MR) imaging have led to the generation of high resolution 3D images of bone structure that correlate with other techniques such as computed tomography (12,13). In addition, MR can quantify the amount of intra-abdominal fat, and MR spectroscopy can also estimate the fat that is present within the bone marrow (14). There is, therefore, the potential to combine these MR-based techniques to obtain objective data on bone microarchitecture and fat content and study these in a condition such as T1DM.

With the increased reports of an association between marrow adiposity and bone health (15), the current study was designed to improve the understanding of the bone pathology in adults with childhood-onset T1DM by using high resolution MRI and biochemical markers of GH action and bone turnover. To reduce the confounding effect of sex hormones, this study targeted young women with T1DM and compared them to a group of age-matched healthy women.

Research Design & Methods

Subjects

Between July 2012 and July 2013, 61 eligible women between the ages of 20 and 30 years and who were diagnosed before the age of 16 years were approached at one hospital clinic and from this group, 30 volunteered to participate. In addition, 28 age-matched healthy control women working at the local university and hospital were also recruited. Exclusion criteria included the presence of metallic implants and pacemakers, active or planned pregnancy or lactation, kidney disease, chronic use of drugs that are known to affect bone health and other chronic diseases that are known to be associated with an increased risk of fractures. Information on personal health and lifestyle habits, including cigarette smoking, alcohol consumption, current medication, use of vitamins or calcium, age at menarche, use of oral contraceptives, hours of weight-bearing physical activity per week, history of fractures and a family history of early osteoporosis was also collected. Information on age of diagnosis, disease duration, insulin therapy and presence of microvascular complications was obtained from the case records. A glycosylated hemoglobin (HbA1c) measurement within a two-week period of the scan visit was used as current HbA1c. The study protocol was approved by the national research ethics service and all participants provided written informed consent.

Biochemical Markers of Bone Metabolism, Adiposity & GH/IGF-1 Axis

In 25 cases and 24 control participants, non-fasting blood samples were collected, centrifuged and the supernatant stored at -80°C . Sample collection was standardised with collection performed in the afternoon to coincide with the clinic visit. Plasma total osteocalcin (OC) and serum bone-specific alkaline phosphatase (BAP), were analyzed by ELISA to assess bone formation (Immunodiagnostic systems, Boldon, Tyne and Wear, United Kingdom). Plasma undercarboxylated osteocalcin (uOC) was used to assess the inactive form of osteocalcin (Cusabio Life Science, Wuhan, P.R. China) and plasma C

terminal telopeptide of Type I collagen (CTX) was measured by ELISA to assess bone resorption (Immunodiagnostic systems, Boldon, Tyne and Wear, United Kingdom). The intra-assay variation for OC, uOC, BAP and CTX was 5.3%, 13.2%, 0.8% and 3.8%, respectively. Plasma leptin, IGF-I and its binding proteins, IGFBP3, and the acid labile subunit (ALS) were also determined, using ELISA (Mediagnost GmbH, Reutlingen, Germany). Intra-assay variabilities were 6.1%, 0.6%, 4.1% and 3.4%, respectively. 25-Hydroxy vitamin D (25OHD) concentration was measured by a radioimmunoassay (Immunodiagnostic Systems, Boldon, UK).

Micro-MRI

MRI images of the proximal tibia with a resolution of 0.3mm x 0.3mm x 0.3mm were acquired with a 3T MRI scanner (Siemens Verio, Erlangen Germany) using a transmit/receive extremity coil which was suitable for knee imaging. The method used has been described previously (16) but briefly, the images were acquired from the right proximal tibia using the epiphyseal growth plate as a reference, with the first slice positioned immediately distal to the growth plate and subsequent slices positioned distally along the tibia. Standardized analysis was performed using the slice that was located at the insertion point of the patellar ligament. The images (Fig.1) were coded and analysed blindly using software written in IDL (Research Systems Inc, Boulder, CO) to obtain measures for apparent bone volume to total volume ratio (appBV/TV), apparent trabecular number (appTbN), apparent trabecular thickness (appTbTh) and apparent trabecular spacing (appTbSp). The analysis for each subject was repeated 4 times and averaged. In 29 out of 30 cases and in 27 out of 28 control participants, the images were of a sufficiently good quality to be analysed. Validation of the software was performed using a custom-made phantom, which consisted of a cylinder containing two discs with a parallel arrangement of nylon strings – 0.4-mm diameter string was used in one disc and 0.2-mm diameter string in the other. In both cases, the strings were split into three regions

with different spacings in each – 0.5, 1.0 and 1.5 mm. The phantom was filled with a NiSO₄ solution to ensure adequate signal surrounding the strings, in a similar way to soft tissue surrounding bone. Due to partial volume effects and the similar magnitude of the resolution of the micro-MRI sequence to the thickness of the strings, the exact thickness of the strings could not be measured, and hence, the calculated bone parameters were expressed as ‘apparent’ values. The phantom validation allowed an optimal assessment of the effect of string thickness and string spacing on the apparent values. An assessment of repeatability was performed by one operator who independently analysed ten datasets four times each.

MRS

¹H-MRS was performed using a 6-channel body array (anterior) and a 12-channel spine coil (posterior). Spectra were obtained from a 20mm x 20mm x 20mm volume within the vertebral body of L3, using a method which has been described previously (16). Analysis was performed following fitting of the spectrum in the time domain using a nonlinear least-squares algorithm, AMARES (17) in the Java-based magnetic resonance user interface (jMRUI) software package (18). The area under the water peak and lipid peak (Fig.2) were obtained and used to calculate the lipid to water ratio (LWR), then percentage fat fraction (%FF) was calculated using the following equation (19):

$$\%FF = \left(\frac{LWR}{LWR + 1} \right) \times 100$$

MRS data were available in all 30 cases and 28 controls.

MRI of abdominal fat

Of the participants who had MRS scans, in 24 cases and 19 controls, approval was also obtained to assess abdominal fat during the same scanning session using the 6-channel body array and the 12-channel spine coil. A T1 weighted turbo-spin-echo sequence was

used to acquire 5 axial slices at the level of L3 with a 4 mm slice thickness using the following parameters: (TR) Repetition Time 300ms, (TE) Echo Time 11ms, (TA) Acquisition Time 17.2s, matrix size=288x320, with 255×340 mm field of view. The images were analysed with Slice-O-matic™ (version 4.3, Tomovision, Canada) for semi-automatic measurement of cross-sectional area (CSA), subcutaneous adipose tissue, (SAT), visceral adipose tissue (VAT) and total adipose tissue (TAT).

Calculation of Sample Size & Statistical Analysis

The primary hypothesis in the study was that cases with T1DM would have a lower appBV/TV and appTbN and a higher appTbSp compared to controls. Based on recent studies from our group (16), the estimated CV of the microMRI measurements was less than 5%. To show a 10% difference between case and controls with a significant difference at $p < 0.05$ with a power of 0.8, at least 16 cases and 16 controls were required for the above microMRI parameters. Data analysis was performed using XLSTAT v2013.3.01 (Addinsoft, Paris, France). All data were described as medians and ranges; comparison between the cases and controls was performed, initially by the Mann-Whitney U test for continuous variables and by the Chi Squared test for categorical variables and then subsequently adjusted for multiple comparisons using False Discovery Rates (FDR) (20). Univariate analysis between continuous variables was performed using the Pearson correlation coefficient.

Results

Clinical Characteristics

There were no significant demographic or anthropometric differences between the cases and controls (Table 1). Notably, the median BMI in the cases and controls was 24.8 kg/m^2

(18.2, 31.2) and 22.9kg/m² (18.3, 33.9), respectively (p=0.1) with similar amounts of physical activity reported in the cases and controls. Of the 30 cases and 28 controls, 9 (30%) and 3 (11%) reported a history of traumatic fractures (p=0.07). In 2 cases, the fractures had occurred before the onset of diabetes and in the other 7 cases, the duration of diabetes at the time of the fracture ranged between 2.5yrs and 16yrs. The median duration of T1DM was 12.8yrs (7.9, 34.2) with a median age at diagnosis of 9.9yrs (0.5, 15.0). Of the 30 cases, 7 were on twice-daily insulin injections, 6 were on three-daily injections and 17 were on four-daily injections. The median total daily insulin dose for body weight was 0.94 IU/kg/d (0.43, 1.52) and the median current HbA1C was 9.8% (5, 16) equivalent to 84mmol/mol (31, 151). Of the 30 cases, 15 (50%) had retinopathy of which 12 had background retinopathy alone, 2 (7%) were being treated for hypertension, 1 (3%) had neuropathy and gastroparesis and none had microalbuminuria. One of the cases had stable Crohn's disease requiring sulfasalazine therapy only for two years prior to the study and two cases were on a stable dose of thyroxine for acquired hypothyroidism, one of three years duration and another of five years duration.

Biochemical Markers of Bone Metabolism, Adiposity & GH/IGF-1 Axis

Amongst the markers of bone metabolism, plasma 25OHD and CTX were significantly lower in cases compared to controls (Table 1). However, serum PTH and markers of bone formation were similar in both groups (Table 1). Amongst the markers of the GH/IGF-1 axis, plasma IGF-1 and ALS were significantly lower in the cases compared to controls whilst IGFBP3 was similar in both groups (Table 1). Plasma leptin was similar in the two groups and, as expected, showed a positive association to BMI (r,0.33; p=0.02) (Table 1). There was no association between HbA1c or the presence of retinopathy with the markers of bone metabolism, GH/IGF-1 axis or leptin.

MRI of abdominal fat

Despite no significant differences in the BMI and plasma leptin between cases and controls, abdominal adiposity including CSA, VAT and TAT, was significantly higher in cases compared to controls (Table 2). However, SCAT was less markedly higher in cases and the difference did not reach statistical significance. Serum leptin did not show an association to any of the MRI-based markers of abdominal obesity. In addition, there was no association between adiposity with markers of control, markers of bone metabolism, GH/IGF-1 axis or leptin.

Bone Microarchitecture by Micro-MRI

Comparison of bone microarchitecture variables revealed that appBV/TV and appTbN were significantly lower and appTbSp significantly higher in cases compared to controls (Table 2). AppBV/TV and appTbN did not show any association to any biochemical markers or MRI-based markers of abdominal obesity. However, appTbSp showed a significant inverse association to serum ALS ($r=-0.30$, $p=0.04$) and OC ($r=-0.38$, $p=0.009$) (Fig.3). This inverse association was not observed between appTbSp and uOC. An inverse association was also suggested between appTbSp and serum IGF-1 but this did not reach statistical significance ($r=-0.27$, $p=0.06$). In the T1DM cases, there was no association of appBV/TV, appTbN or app TbSp with HbA1c, age at diagnosis, duration of T1DM or daily insulin dose corrected for weight. However, there was a clear difference in median appBV/TV between those cases who had retinopathy and who did not have retinopathy. The median appBV/TV was 0.285 (0.22, 0.33) in those cases with retinopathy compared to 0.33 (0.25, 0.37) in those cases without retinopathy ($p=0.02$) (Fig.4). The HbA1c was 9.5% (5, 16) and 9.2% (5.5, 13) in the cases with and without retinopathy (NS).

Bone Marrow Adiposity

Median vertebral BMA, expressed as percentage fat fraction (%FF), was higher in cases than controls but this did not reach statistical significant difference (Table 2). Vertebral BMA tended to show a positive association to VAT ($r=0.3$, $p=0.051$) which, as stated earlier, was higher in cases compared to controls (Table 2). There was no association between vertebral BMA and markers of GH/IGF-1 axis or bone turnover. Median BMA was 30.5% (9.9, 59.9) in those cases with retinopathy compared to 31.6% (11.3, 50.1) in those cases without retinopathy ($p=0.97$).

Discussion

The current study was primarily aimed at using MRI to compare the trabecular microarchitecture in young women with childhood-onset T1DM with that in healthy age-matched women and is the first published clinical study to clearly show that this is altered in the former with reduced bone volume and trabecular number and increased trabecular separation. In addition, the study has also provided some insights into the underlying mechanisms that may lead to abnormality of skeletal development in adults with T1DM.

In keeping with other reports of a higher prevalence of vitamin D deficiency in young people with T1DM, the women who participated in this study had lower vitamin D levels than the control group (21, 22). However, these levels were not particularly low and were not associated with a raised PTH or any other marker of bone formation or bone microarchitecture and it is, therefore, unclear whether the the lower vitamin D levels had a particularly marked contributory role. It is also unlikely that the women with T1DM had any other underlying genetic bone disorder as none had been revealed on history and examination. The reported rate of fractures in these cases seemed higher but not significantly different to controls and within the expected frequency for self-reported fractures in healthy women (23).

The findings in the current study suggest that the deficits in bone microarchitecture that were encountered in T1DM may have a multifactorial aetiology. Firstly, the negative association between total osteocalcin, a marker of bone formation and trabecular separation in the study participants suggests that the osteopathy in T1DM may be linked to reduced bone formation rather than increased bone resorption. This was further reinforced by the lower levels of CTX, a marker of bone resorption, in the T1DM cases. This state of low bone turnover with reduced bone formation in T1DM has been reported previously (24, 25) and has been known to be associated with GH deficiency (26). To

facilitate recruitment, the current study did not stipulate fasting blood samples. Markers of bone turnover, and specially markers of bone resorption, such as CTX, show a diurnal variation and may be affected by fasting status (27) and future studies would benefit from standardisation of sample collection.

The women with T1DM also had lower levels of circulating IGF-1 and ALS and the negative association of trabecular separation with ALS raises the possibility that the GH/IGF-1 axis and, in particular, reduced GH activity, may contribute to the altered bone microarchitecture. GH and IGF-1 are important regulators of bone homeostasis and important for the maintenance of bone mass (7). GH stimulates osteoblastic proliferation through stimulation of circulating and local IGF-1 and it inhibits adipogenesis (28). In humans, systemic availability of active IGF-1 is modulated through its binding to proteins such as IGFBP-3 and ALS (29), which, along with IGF-1, are primarily synthesized in the liver under the stimulation of GH (30). Insulin deficiency is reported to be associated with a state of GH resistance (31) an abnormality of post-translational synthesis of ALS and ALS deficiency (32, 33) and with lower levels of ALS, circulating IGF-1 falls due to increased renal clearance. As the insulin deficiency in the portal circulation exists despite insulin replacement in T1DM (34), it is likely that this state of GH resistance is not alleviated during treatment in T1DM and the abnormal bone findings in this study may be a consequence of a persistent state of IGF-1 and ALS deficiency. The finding in the current study of an inverse association between trabecular separation and serum osteocalcin and ALS reinforces this view.

The other possible reason for the abnormality in bone microarchitecture in T1DM may be related to microvasculopathy. The clear association of reduced bone volume in those cases of T1DM who had retinopathy, albeit background retinopathy in most cases, suggests that it is possible that the underlying pathophysiology of retinopathy may also

influence trabecular development. The pathology of diabetic retinopathy evolves in an environment of increased formation of reactive oxygen species, leukostasis and breakdown of the blood-retinal barrier that is followed by formation of acellular capillaries and development of micro aneurysms (35). The superoxide generating family of NADPH oxidase enzymes have not only been strongly implicated in the vascular complications of diabetes but superoxide radicals may also be associated with abnormalities of bone turnover (36, 37). The possibility of a microvasculopathy that affects bone in T1DM has not been studied widely and requires further exploration.

The women with T1DM in this study did not have a significantly higher BMI compared to controls but they had markedly higher visceral adiposity and it is possible that this abnormality is also associated with functional GH deficiency (38). Although bone marrow adiposity was similar in the cases and controls, the cases did have increased trabecular spacing which showed an inverse relationship to circulating ALS and osteocalcin. Increased trabecular spacing is not universally associated with increased bone marrow adiposity (15); whereas this association may be encountered in GH deficiency (16, 39), in other conditions such as osteogenesis imperfecta, the two parameters do not show an association (16). It is also possible that in states of GH resistance such as T1DM, bone marrow adiposity may be increased. The current study was not powered to detect significant differences in bone marrow adiposity between the cases and controls and it is possible that with a larger sample size this would also have been detected. The finding of an abnormality of bone microarchitecture and bone marrow adiposity would be consistent with a defect in bone marrow mesenchymal stem cell differentiation which leads to a net reduction in bone formation in favour of adipogenesis (40) and as previously described in streptozocin induced T1DM mice (41) as well as conditions that are associated with relative IGF-1 deficiency (8,42,43).

375 Finally, the current study highlights the objective versatility of MRI based techniques in
376 identifying differences in relatively small cohorts. Although there is now sufficient
377 supporting evidence for the use of MRI for assessing bone microarchitecture (44), with
378 the greater availability of 3T MRI scanners, there is a need to standardise the analytical
379 techniques so that the clinical utility of MRI in metabolic conditions that are associated
380 with abnormalities of bone health can be realised. With technological advances in
381 hardware as well as analytical tools, an improvement in the current limited resolution of
382 MRI as well as its ability to study cortical structure is highly likely. We chose to study
383 young women only, primarily because they are more likely to develop osteoporosis in the
384 long-term and to avoid the confounding effects of sex hormones but given that the
385 increased fracture incidence has been encountered in both sexes and bone marrow
386 adiposity increases with old age (45, 46), there is a need to study older as well as younger
387 people with diabetes in both sexes. We did not perform DXA in this group but there is a
388 need in future studies to assess the relationship of microMRI findings to DXA BMD as
389 well as DXA based methods of assessing trabecular architecture (47). Given that hip
390 fractures are reported to be commoner in people with T1DM (1,2,11), there is a place to
391 examine the microarchitecture at this site too. The women with T1DM in this current
392 group had a relatively high HbA1c and as expected for a population sample from an inner
393 city area in the west of Scotland (48). A relationship between HbA1c and the bone
394 parameters was not observed in the current study and perhaps this investigation would
395 have been facilitated if there was a wider range of HbA1c. The lack of a relationship of
396 the bone findings with HbA1c also raises the need to investigate the association with
397 additional markers of glycation such as pentosidine (49). The lack of a sufficient number
398 of cases with other complications associated with microvasculopathy limited the
399 investigation of this association in the current study. The relationship between bone
400 pathology and Type 2 DM is less clear with a lower risk of fractures (3). However, a
401 recent study that examined bone marrow adiposity in women with Type 2 DM did report

a higher level of vertebral bone marrow adiposity in those who had a history of a fracture (50).

In summary, detailed MRI studies in young women with T1DM have shown clear abnormalities of bone health that are characterised by reduced trabecular bone volume and reduced trabecular numbers. Possible underlying mechanisms that require further exploration include a microvasculopathy and GH resistance.

Disclosures

The authors do not have any relevant conflicts of interest.

Acknowledgements

We would like to acknowledge the support of the Medical Fund of the University of Glasgow. We would also like to thank Tracey Steedman, Kirsten Lanaghan, Heather Boylan, Alison Frood and Evonne McLennan for their assistance with image acquisition. NA is supported by the Government of Libya. CM is supported through the Scottish Academic Health Sciences Collaboration. NA recruited the cases, performed the study, analysed the data and wrote the manuscript, CM performed the study, analysed the data and wrote the manuscript, JF analysed the data and revised the manuscript, RSL designed the study, recruited the cases and revised the manuscript⁴, JM analysed the data and revised the manuscript, MM performed the assays and revised the manuscript, RD, DG, GM, CP recruited the cases and revised the manuscript, MGS designed the study and revised the manuscript; SFA conceived and designed the study, performed the analysis, wrote the manuscript and acts as the guarantor.

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Legend To Figures

Figure 1 Representative coronal 3T MR image of the proximal tibia. (A) showing the placement of the reference line and the region of interest captured in the axial images. (B) Micro-MR images. (C) Semiautomatic segmentation delineates the trabecular bone compartment. (D) The region of interest (ROI) is binarised to produce an image of a pure bone phase and a pure marrow phase. (E) Higher magnification view of section within the ROI. (F) A 3-D reconstruction of the trabecular bone within the ROI.

Figure 2 Typical ^1H -magnetic resonance spectrum of vertebral body L3. Bottom panel: original data. Middle panel: estimated spectrum fit as performed by the Java based Magnetic Resonance User Interface (jMRUI) software package. Top: individual peak components as fitted with jMRUI. Peak definitions are: 1 – water; 2 – lipid.

Figure 3 The relationship between apparent trabecular separation (appTbSp) as assessed by MRI and serum acid labile subunit (ALS, panel A) and serum osteocalcin (OC, panel B) in women with T1DM (filled circles) and controls (open circles). A Gaussian ellipse of the data has been added to each plot, together with the correlation (r) and its p-value (p). An inverse association between appTbSp and ALS and OC was observed.

Figure 4 Apparent bone volume/total volume (appBV/TV) in women with T1DM with and without retinopathy and compared to control women (open circles).

	T1DM	Controls	
N	30	28	p
Age (yrs)	22.0 (16.9, 36.1)	21.8 (18.6, 37.5)	<i>0.7</i>
BMI	24.8 (18.2, 31.2)	22.9 (18.3, 33.9)	<i>0.1</i>
Cigarette smoker (n)	4	0	<i>0.05</i>
Regular alcohol consumption (n)	25	27	<i>0.9</i>
Ca, Vit D, Multivit supplements (n)	4	6	<i>0.8</i>
Age at menarche (yrs)	12 (10, 17.5)	13 (11, 15)	<i>0.6</i>
Previous pregnancy (n)	3	1	<i>0.3</i>
Oral contraceptive use (n)	21	22	<i>0.9</i>
Physical activity (hrs/wk)	4 (0, 10)	4 (0, 10)	<i>0.9</i>
PTH (pmol/l)	3.4 (1.1,9)	4.2 (1.4,12.5)	<i>0.3</i>
25OH D (nmol/l)	33 (19, 65)	45.5 (25, 84)	<i>0.02</i>
OC (µg/l)	13.4 (3.6, 20.8)	14.3 (6.6, 40.5)	<i>0.2</i>
<u>uOC</u> (ng/ml)	<u>1.8 (0.5, 13.9)</u>	<u>3.3 (0.5, 25.3)</u>	<u><i>0.18</i></u>
Bone ALP (U/l)	16.2 (10.5, 44.3)	17.2 (4.6, 28.8)	<i>0.9</i>
CTX (µg/l)	0.15 (0.03, 0.39)	0.20 (0.05, 0.49)	<i>0.04</i>
IGF-1 (µg/l)	135.2 (66.5, 83.1)	182.4 (83.1, 293.4)	<i>0.002</i>
IGFBP-3 (µg/l)	2208 (1794, 3677)	2347 (1991, 3538)	<i>0.17</i>
ALS (mU/ml)	2064 (1005, 3009)	2549 (1929, 3682)	<i><0.0001</i>
Leptin (µg/l)	12.4 (1.3, 105)	14.3 (2.6, 43.3)	<i>0.96</i>

Table 1 – Clinical and biochemical details of study participants. All continuous variables are described as median and range. PTH, Parathyroid hormone; OC, osteocalcin; uOC, undecarboxylated osteocalcin; 25OHD, 25-hydroxyvitamin D; ALP, Alkaline phosphatase; CTX, crosslinked C-terminal telopeptides of type I collagen; IGF-1, insulin-like growth factor 1; IGFBP-3, insulin-like growth factor binding protein 3; ALS, acid labile subunit. Significance is assigned at $p < 0.05$.

	T1DM	Controls	<i>P</i>*
N	30	28	
CSA (mm ²)	47,878 (30,134, 78,651)	39,918 (30,232, 69,789)	<i>0.03</i>
SCAT (mm ³)	21,583 (3,719, 42,877)	15,582 (7,454, 40,807)	<i>0.068</i>
VAT (mm ³)	5,733 (2,030, 11,144)	3,460 (1,808, 6,832)	<i>0.012</i>
TAT (mm ³)	27,230 (6,584, 54,021)	19,130 (10,046, 47,639)	<i>0.03</i>
AppBV/TV	0.3 (0.22, 0.37)	0.33 (0.26, 0.4)	<i>0.018</i>
AppTb.N(mm-1)	0.26 (0.2, 0.3)	0.29 (0.23, 0.3)	<i>0.012</i>
AppTb.Sp(mm)	2.59 (2.24, 3.38)	2.32 (2.03, 2.97)	<i>0.012</i>
AppTb.Th(mm)	1.14 (0.86, 1.49)	1.15 (0.96, 1.39)	<i>0.3</i>
L3 fat fraction (%)	31.1 (9.9, 59.9)	26.3 (8.5, 49.8)	<i>0.2</i>

Table 2 MRI-based measures of abdominal adiposity, bone microarchitecture and vertebral bone marrow adiposity at L3. All data are described as median and range. CSA- cross-sectional area of adipose tissue; SCAT – subcutaneous adipose tissue; VAT- visceral adipose tissue; TAT – total adipose tissue; AppBV/TV – apparent bone volume/total volume; AppTb.N – apparent trabecular number; AppTbSp – apparent trabecular separation; AppTbTh – apparent trabecular thickness. P-values have been adjusted for multiple comparisons using False Discovery Rates; significance has been assigned at adjusted p<0.05.

Fig.1

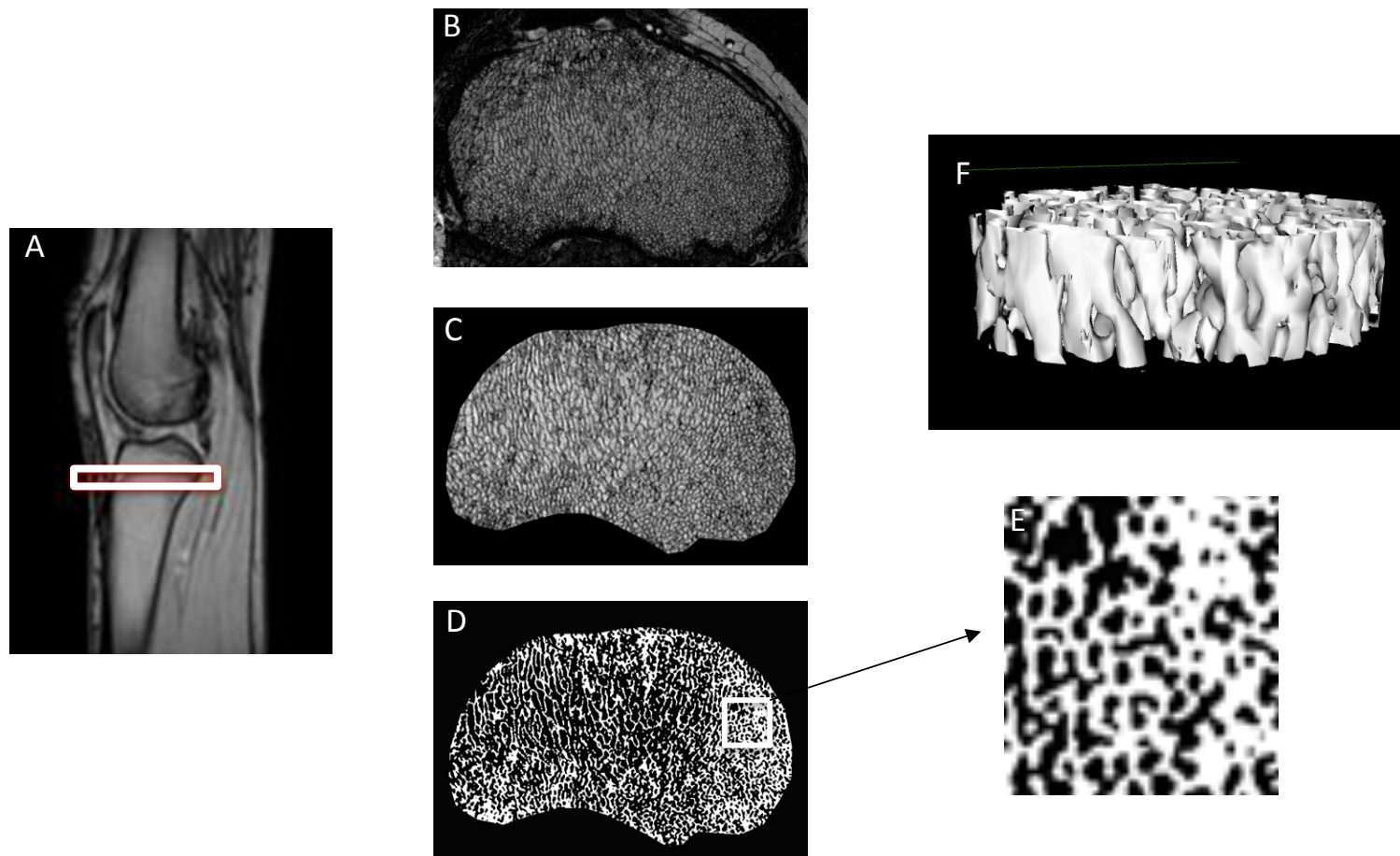


Fig.2

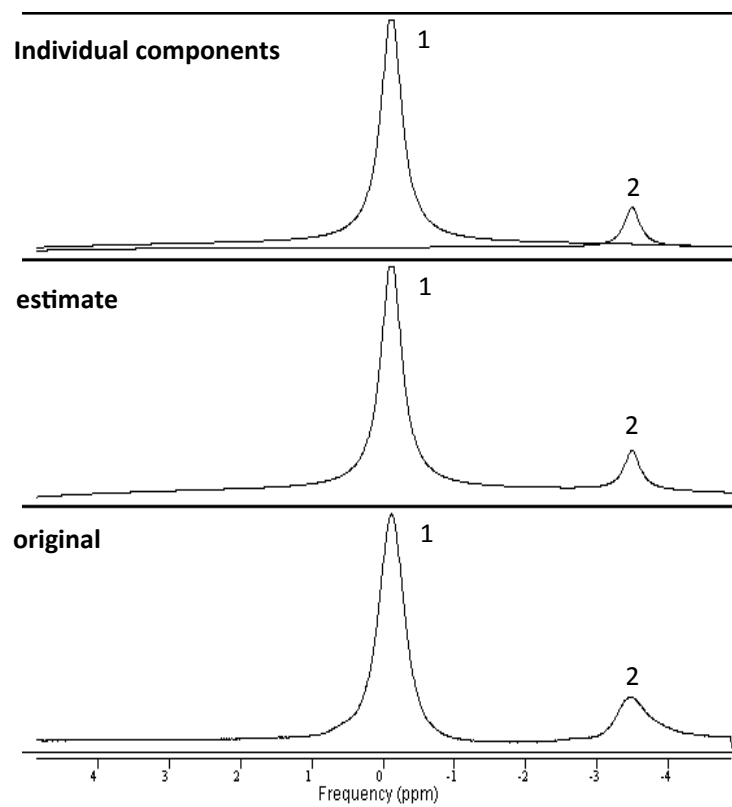


Fig.3

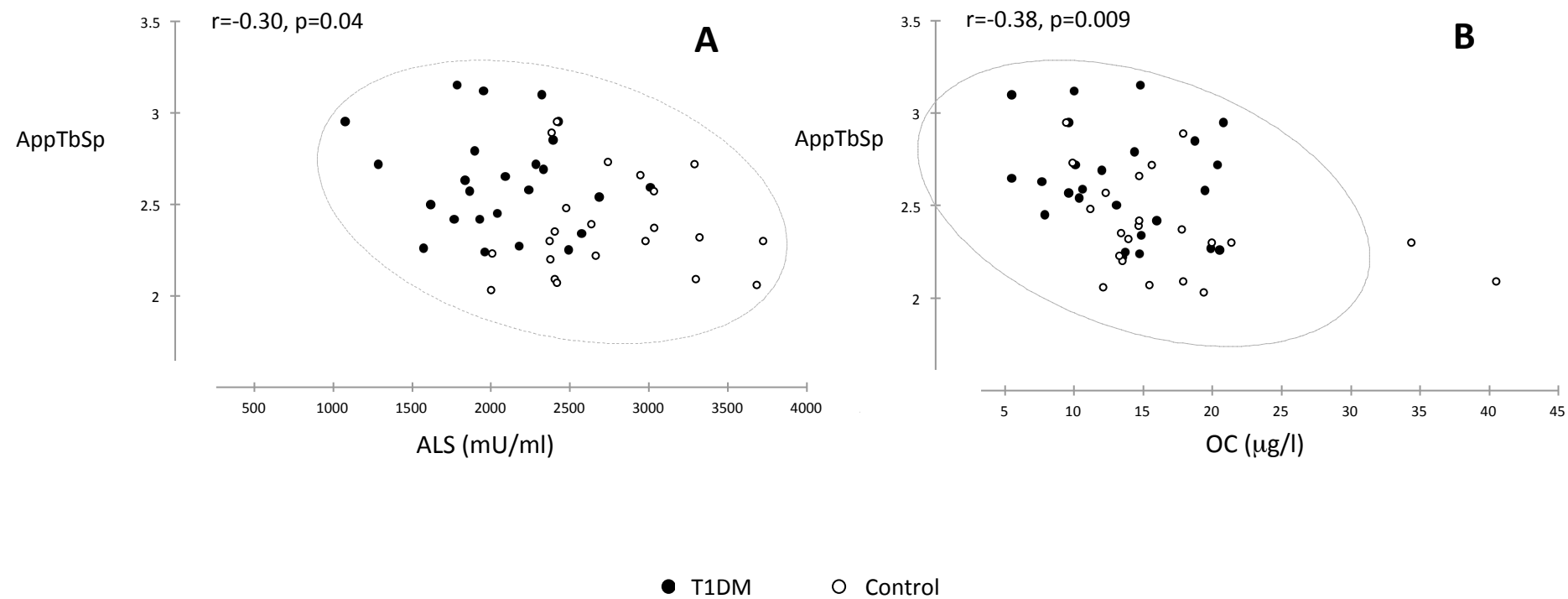


Fig.4

